(For Laboratory Use Only)
Accuratus Lab Services Project # A19691 CWK 11/25/15

ACCURATUS LAB SERVICES

PROTOCOL

Standard Test Method for Efficacy of Sanitizers Recommended for Soft Non-Food Contact Surfaces (Dilutable)

Test Organism(s):

Staphylococcus aureus (ATCC 6538)
Klebsiella pneumoniae (ATCC 4352)

PROTOCOL NUMBER

SRC85090915.NFS.1

PREPARED FOR/SPONSOR

Church & Dwight Co., Inc.
Corporate Technical Center
469 North Harrison Street
Princeton, NJ 08543

SPONSOR REPRESENTATIVE

Scientific & Regulatory Consultants, Inc.
201 W. Van Buren Street
Columbia City, IN 46725

PREPARED BY/TESTING FACILITY

Accuratus Lab Services
1285 Corporate Center Drive, Suite 110
Eagan, MN 55121

DATE

September 9, 2015

Revised November 16, 2015

EXACT COPY
INITIALS MAT DATE 1/12/16

Standard Test Method for Efficacy of Sanitizers Recommended for Soft Non-Food Contact Surfaces (Dilutable)

PURPOSE

The purpose of this study is to determine the antimicrobial efficacy of sanitizers on soft, non-food contact surfaces. This method is in compliance with the requirements of and may be submitted to, one or more of the following agencies as indicated by the Sponsor: U.S. Environmental Protection Agency (EPA) and Health Canada.

TEST SUBSTANCE CHARACTERIZATION

According to 40 CFR, Part 160, Subpart F [160.105] test substance characterization as to identity, strength, purity, solubility and composition, as applicable, shall be documented before its use in this study. The stability of the test substance shall be determined prior to or concurrently with this study. Pertinent information, which may affect the outcome of this study, shall be communicated in writing to the Study Director upon sample submission to Accuratus Lab Services. Accuratus Lab Services will append Sponsor-provided Certificates of Analysis (C of A) to this study report, if requested and supplied. Characterization and stability studies not performed following GLP regulations will be noted in the Good Laboratory Practice compliance statement.

SCHEDULING AND DISCLAIMER OF WARRANTY

Experimental start dates are generally scheduled on a first-come/first-serve basis once Accuratus Lab Services receives the Sponsor approved/completed protocol, signed fee schedule and corresponding test substance(s). Based on all required materials being received at this time, the proposed experimental start date is December 4, 2015. Verbal results may be given upon completion of the study with a written report to follow on the proposed completion date of January 4, 2016. To expedite scheduling, please be sure all required paperwork and test substance documentation is complete/accurate upon arrival at Accuratus Lab Services.

If a test must be repeated, or a portion of it, due to failure by Accuratus Lab Services to adhere to specified procedures, it will be repeated free of charge. If a test must be repeated, or a portion of it, due to failure of internal controls, it will be repeated free of charge. "Methods Development" fees shall be assessed, however, if the test substance and/or test system require modifications due to complexity and difficulty of testing.

If the Sponsor requests a repeat test, they will be charged for an additional test. Neither the name of Accuratus Lab Services nor any of its employees are to be used in advertising or other promotion without written consent from Accuratus Lab Services. The Sponsor is responsible for any rejection of the final report by the regulating agencies concerning report format, pagination, etc. To prevent rejection, Sponsor should carefully review the Accuratus Lab Services final report and notify Accuratus Lab Services of any perceived deficiencies in these areas before submission of the report to the regulatory agency. Accuratus Lab Services will make reasonable changes deemed necessary by the Sponsor, without altering the technical data.

JUSTIFICATION FOR SELECTION OF THE TEST SYSTEM

The U.S. Environmental Protection Agency requires that a specific claim for a sanitizer be supported by appropriate scientific data demonstrating the efficacy of the sanitizer against the claimed organism. In addition, Health Canada requires that the product be recognized as a disinfectant prior to accepting sanitizer claims. This is accomplished in the laboratory by treating the target organism with the test substance under conditions which simulate as closely as possible, the actual conditions under which the test substance is designed to be used. For products intended for use on soft non-food contact surfaces, a fabric carrier method is used in the generation of the supporting data. The test system to be used in this study will be a modification of the ASTM approved method for the evaluation of the antimicrobial efficacy of sanitizers on soft non-food contact surfaces. The carriers and associated drying conditions have been modified for a soft-surface application.

TEST PRINCIPLE

A film of organism cells dried on a surface of appropriate carriers is exposed to the test substance for a specified exposure time. After exposure, the carriers are neutralized and assayed for survivors. Appropriate sterility, culture purity, carrier population, neutralization confirmation and inoculum count controls are performed. The current revision of Standard Operating Procedure CGT-0032 reflects the methods which shall be used in this study.

TEST METHOD

Table 1:

Test Organism	Designation #	Growth Medium	Incubation Parameters
Staphylococcus aureus	6538	Nutrient Broth	35-37°C, aerobic
Klebsiella pneumoniae	4352	Nutrient Broth	35-37°C, aerobic

The test organism(s) to be used in this study was/were obtained from the American Type Culture Collection (ATCC), Manassas, VA.

Subculture Agar: Tryptic Soy Agar+5% Sheep's blood will be used in testing. The agar used in the test will be the same as that which is used in the control procedures which substantiates test organism recovery.

Carriers

Prepare a scouring solution by adding a ratio of approximately 1.5 grams Na_2CO_3 and approximately 1.5 grams of Triton X-100 to approximately 3 L of deionized water. Equivalent dilutions may be made. The fabric for testing will be obtained containing approximately 80 x 80 threads/inch, plain cotton weave. Alternate fabric may be used by Sponsor request. Add approximately 300 grams of test fabric to each 3 L volume of scouring solution, or equivalent. Allow the solution to boil for approximately 60 minutes. Remove the fabric and rinse by placing first in boiling water for a minimum of 5 minutes and then placing in cold water for a minimum of 5 minutes. During the rinsing procedure, mix the fabric in the water in order to help remove wetting agent. Allow the fabric to air dry. Cut fabric carriers of approximately 1 inch x 1 inch from the prepared fabric and autoclave sterilize. After sterilization, place each carrier into a sterile Petri dish prior to use in testing.

Preparation of Test Organism

From a stock slant no more than 5 transfers from original stock and ≤1 month old, an initial tube (10 mL) of culture broth will be inoculated. This culture is termed the "initial broth suspension." From this initial broth suspension, at least three consecutive daily transfers using 1 loopful (10 µL) of culture into 10 mL of culture media will be performed prior to use as an inoculum. Incubate each daily transfer for 24±2 hours using the appropriate growth medium. The final test culture will be incubated for 48-54 hours.

A 48-54 hour culture will be vortex-mixed and allowed to settle for ≥15 minutes. The upper 2/3rds of the culture will be removed and transferred to a sterile vessel for use in testing. The culture may be adjusted by dilution in growth medium or by centrifuge concentration, if necessary. An organic soil load may be added to the test culture per Sponsor request. The test culture will be thoroughly mixed prior to use.

Preparation of Test Substance

The test substance will be prepared according to the directions for intended use of the product. The test substance shall be used within three hours of preparation if additional preparation is required by Accuratus Lab Services.

Contamination of Carriers
Inoculate each sterile carrier with 0.03 mL (30 µL) of culture using a calibrated pipettor distributing the inoculum evenly about the carrier. Dry the inoculated carriers for 20-30 minutes until visibly dry with the Petri dish lids intact. A drying humidity should be selected to encourage maximum survival of the test organism (targeting approximately 40% humidity, for example). A constant humidity chamber will be used in place of a desiccating chamber to ensure uniform humidification conditions and to overcome slow re-equilibration of a desiccator after opening.

Drying Conditions: 35-37°C targeting 40% humidity

Exposure Conditions
Following the completion of drying, transfer each carrier to individual sterile 2 oz. (60 mL) polypropylene jars using sterile forceps. Using staggered intervals, transfer 5.0 mL of prepared test substance to each jar. The liquid should completely cover the carrier during exposure. Allow the carriers to expose at the Sponsor specified exposure temperature for the Sponsor specified exposure time. Following exposure, transfer 20 mL of neutralizer to the jars using identical staggered intervals. **Vortex mix the jars for 10-15 seconds.** Glass beads may be utilized to aid in organism recovery from the modified fabric substrate.

Test System Recovery
Within 30 minutes of neutralization, plate 1.0 mL and 0.1 mL aliquots of the neutralized subcultures (10^0) in duplicate onto appropriate agar.

If neutralization of the test substance cannot be achieved chemically, filter-neutralization may be performed. Within 30 minutes of neutralization, transfer duplicate 1.0 mL and 0.1 mL of the neutralized solution, to individual filter units pre-wetted with 10 mL of sterile diluent. Evacuate the contents and rinse each filter with a minimum of 50 mL of sterile diluent. Transfer each filter to an appropriate agar using sterile forceps.

Incubation and Observation
All subcultures are incubated under the conditions listed in table 1 for 48±4 hours. Following incubation, the subcultures will be visually enumerated. If necessary, the subcultures may be placed at 2-8°C for up to three days prior to examination.

Representative test plates showing growth may be subcultured, stained and/or biochemically assayed to confirm or rule out the presence of the test organism. If possible, subcultures containing 30-300 colonies will be used for calculations. When membrane filtration is used, the upper limit used for counting/calculations should be 200 CFU.

STUDY CONTROLS

Carrier Population Control
Inoculated, dried control carriers will be treated as in a fashion similar to the test procedure utilizing sterile deionized water in place of the test substance. If multiple exposure times were followed in testing, the carriers will be exposed for the shortest exposure time followed in the test procedure. Following exposure, the carriers will be neutralized as in the test. The carriers will be vortex-mixed to ensure complete elution of the test organism. Ten-fold serial dilutions will be prepared and 0.1 mL aliquots of the 10^{-1} to 10^{-4} dilutions will be plated in duplicate. The acceptance criterion for this study control is a minimum geometric mean value of 7.5×10^5 CFU/carrier.

Carrier Sterility Control
Prior to testing, or concurrent with testing, a representative, uninoculated carrier will be added to the neutralizer. The vessel will be mixed and 1.0 mL will be plated onto appropriate agar and incubated. The acceptance criterion is a lack of growth following incubation.

Neutralizer Sterility
Prior to or concurrent with testing, a 1.0 mL aliquot of neutralizer will be plated onto appropriate agar and incubated. The acceptance criterion is a lack of growth following incubation.

Culture Purity
A "streak plate for isolation" will be performed on the organism culture and following incubation examined in order to confirm the presence of a pure culture. The acceptance criterion for this study control is a pure culture demonstrating colony morphology typical of the test organism.

Organic Soil Load Sterility
Prior to or concurrent with testing and if applicable, the serum used for the organic soil load will be cultured, incubated, and visually examined for lack of growth. The acceptance criterion is a lack of growth following incubation.

Neutralization Confirmation Control
In a manner consistent with the AOAC 960.09 method, the following neutralization confirmation control will be performed prior to testing or concurrent with testing. To represent worst-case conditions, only the most concentrated test substance dilution and/or shortest exposure time needs to be utilized in this control when multiple test substance concentrations or multiple exposure times are being evaluated in the study.

Serially dilute the prepared test culture to target $2x10^4 - 2x10^5$ CFU/mL (to target a result of 10-100 CFU plated in each control run). Multiple organism dilutions may be prepared. *(Typically the 10^{-3}, 10^{-4} or 10^{-5} dilutions will provide a culture in range depending on expected titer. Alternate or partial dilutions may be used where appropriate.)* If all the organism dilution(s) used in this control fail to provide adequate numbers which coincides in a failure to meet the acceptance criterion for this study control, the control may be repeated in its entirety.

Test Culture Titer (TCT)
Add 0.1 mL of diluted test organism to 25 mL of sterile diluent and vortex mix. Hold the mixture for a minimum of 30 minutes and spread plate or filter plate duplicate 1.0 mL and 0.1 mL aliquots using the same method used in the test. The acceptance criterion for this study control is growth. If the test culture titer fails to yield countable numbers or if the culture titer is too low resulting in failing results, the entire neutralization confirmation control may be repeated in its entirety, as necessary, to properly validate neutralization.

Neutralization Confirmation Control Treatment (NCT)
Immerse a sterile carrier (one per test organism dilution to be used, per test substance to be evaluated) in 5.0 mL of test substance as in the test. Expose for the exposure time and neutralize each carrier with 20 mL of neutralizer. **Vortex-mix for 10-15 seconds.** Within 5 minutes, add 0.1 mL of diluted test organism to the neutralized contents and vortex mix. Hold the mixture for a minimum of 30 minutes and spread plate or filter plate duplicate 1.0 mL and 0.1 mL aliquots using the same method used in the test. The acceptance criterion for this study control is growth within 1 $\log_{10}$ of the test culture titer (TCT) for at least one of the aliquots plated.

Neutralizer Toxicity Treatment (NTT)
Add 0.1 mL of diluted test organism to 25 mL of sterile neutralizer and vortex mix. Hold the mixture for a minimum of 30 minutes and spread plate or filter plate duplicate 1.0 mL and 0.1 mL aliquots using the same method used in the test. The acceptance criterion for this study control is growth within 1 $\log_{10}$ of the test culture titer (TCT) for at least one of the aliquots plated.

Hold times after the addition of the test organism to the neutralization confirmation control vessels may be reduced if neutralization is a concern. Hold times followed should be as long or longer than the actual time required to plate the test carriers for a given test organism/test substance set.

Inoculum Count
Serially dilute and plate the test organism in duplicate using 0.1 mL aliquots and appropriate dilutions and incubate as in the test. This control is for informational purposes and therefore has no acceptance criterion.

PROCEDURE FOR IDENTIFICATION OF THE TEST SYSTEM

Accuratus Lab Services maintains Standard Operating Procedures (SOPs) relative to efficacy testing studies. Efficacy testing is performed in strict adherence to these SOPs which have been constructed to cover all aspects of the work including, but not limited to, receipt, log-in, and tracking of biological reagents including test organism strains for purposes of identification, receipt and use of chemical reagents. These procedures are designed to document each step of efficacy testing studies. Appropriate references to medium, batch number, etc. are documented in the raw data collected during the course of each study.

Additionally, each efficacy test is assigned a unique Project Number when the protocol for the study is initiated by the Study Director. This number is used for identification of the test subcultures, etc. during the course of the test. Test subcultures are also labeled with reference to the test organism, experimental start date, and test product. Microscopic and/or macroscopic evaluations of positive subcultures are performed in order to confirm the identity of the test organism. These measures are designed to document the identity of the test system.

METHOD FOR CONTROL OF BIAS: N/A

STUDY ACCEPTANCE CRITERIA

Test Substance Performance Criteria
The efficacy performance requirements for label claims state that the test substance must demonstrate a minimum 99.9% reduction of test survivors as compared to the population control to be considered an effective non-food contact sanitizer.

Control Acceptance Criteria
The study controls must perform according to the criteria detailed in the study controls description section. If any control acceptance criteria are not met, the test may be repeated under the current protocol number.

If any portion of the protocol is executed incorrectly warranting repeat testing, the test may be repeated under the current protocol number. If the population control fails to meet the minimum requirement or if the neutralization control acceptance criteria is not met and the study fails to meet the efficacy requirements, repeat testing is not required.

REPORT

The report will include, but not be limited to, identification of the sample, date received, initiation and completion dates, identification of the organism strains used, description of media and reagents, description of the methods employed, tabulated results and conclusion as it relates to the purpose of the test, and all other items required by 40 CFR Part 160.185.

PROTOCOL CHANGES

If it becomes necessary to make changes in the approved protocol, the revision and reasons for changes will be documented, reported to the Sponsor and will become a part of the permanent file for that study. Similarly, the Sponsor will be notified as soon as possible whenever an event occurs that may have an effect on the validity of the study.

Standard operating procedures used in this study will be the correct effective revision at the time of the work. Any minor changes to SOPs (for this study) or methods used will be documented in the raw data and approved by the Study Director.

TEST SUBSTANCE RETENTION

It is the responsibility of the Sponsor to retain a sample of the test substance. All unused test substance will be discarded following study completion unless otherwise indicated by Sponsor.

RECORD RETENTION

Study Specific Documents

All of the original raw data developed exclusively for this study shall be archived at Accuratus Lab Services for a minimum of five years for GLP studies or a minimum of six months for all other studies following the study completion date. After this time, the Sponsor (or the Sponsor Representative, if applicable) will be contacted to determine the final disposition. These original data include, but are not limited to, the following:

1. All handwritten raw data for control and test substances including, but not limited to notebooks, data forms and calculations.
2. Any protocol amendments/deviation notifications.
3. All measured data used in formulating the final report.
4. Memoranda, specifications, and other study specific correspondence relating to interpretation and evaluation of data, other than those documents contained in the final study report.
5. Original signed protocol.
6. Certified copy of final study report.
7. Study-specific SOP deviations made during the study.

Facility Specific Documents

The following records shall also be archived at Accuratus Lab Services. These documents include, but are not limited to, the following:

1. SOPs which pertain to the study conducted.
2. Non study-specific SOP deviations made during the course of this study which may affect the results obtained during this study.
3. Methods which were used or referenced in the study conducted.
4. QA reports for each QA inspection with comments.
5. Facility Records: Temperature Logs (ambient, incubator, etc.), Instrument Logs, Calibration and Maintenance Records.
6. Current curriculum vitae, training records, and job descriptions for all personnel involved in the study.

REFERENCES

1. U.S. Environmental Protection Agency, Office of Chemical Safety and Pollution Prevention, Product Performance Test Guidelines, OCSPP 810.2000: General Considerations for Uses of Antimicrobial Agents, September 4, 2012.
2. U.S. Environmental Protection Agency, Office of Chemical Safety and Pollution Prevention, Product Performance Test Guidelines, OCSPP 810.2400: Disinfectants and Sanitizers for Use on Fabrics and Textiles - Efficacy Data Recommendations, December 21, 2012.
3. American Society for Testing and Materials (ASTM). Standard Test Method for Efficacy of Sanitizers Recommended for Inanimate Non-Food Contact Surfaces, E1153-14.
4. Association of Official Analytical Chemists (AOAC) Official Method 960.09, Germicidal and Detergent Sanitizing Action of Disinfectants Method. In Official Methods of Analysis of the AOAC, 2013 Edition.
5. American Society for Testing and Materials (ASTM). Standard Test Method for Evaluation of Laundry Sanitizers and Disinfectants, E2274-09.
6. Association of Official Analytical Chemists (AOAC) Official Method 961.02, Germicidal Spray Products as Disinfectants. In Official Methods of Analysis of the AOAC, 2012 Edition.
7. Health Canada, January, 2014. Guidance Document – Safety and Efficacy Requirements for Hard Surface Disinfectant Drugs.
8. Health Canada, January, 2014. Guidance Document - Disinfectant Drugs.

DATA ANALYSIS

Calculations

$$\text{CFU/mL} = \frac{\text{(average CFU) x (dilution factor)}}{\text{(volume plated in mL)}}$$

Number of Organisms Surviving per Carrier

$$\text{CFU/carrier} = \frac{\text{(average CFU) x (dilution factor) x (volume neutralized solution in mL)}}{\text{(volume plated or filtered in mL)}}$$

Geometric Mean of Number of Organisms Surviving on Test or Control Carriers

$$\text{Geometric Mean} = \text{Antilog of } \frac{\text{Log}_{10}X_1 + \text{Log}_{10}X_2 + \text{Log}_{10}X_N}{N}$$

Where: X equals CFU/carrier
N equals number of carriers

Percent Reduction

$$\% \text{ reduction} = [(a - b) / a] \times 100$$

where:
a = geometric mean of the number of organisms surviving on the population control carriers.
b = geometric mean of the number of organisms surviving on the test carriers.

Recovery Log_{10} Difference = Log_{10} (Average CFU in TCT) – Log_{10} (Average CFU in NCT or NTT)
Used for the neutralization confirmation control

Statistical Methods
None Used.

STUDY INFORMATION

(All blank sections are completed by the Sponsor or Sponsor Representative as linked to their signature, unless otherwise noted.)

Test Substance (Name & Batch Numbers) exactly as it should appear on final report:

Capricorn Lots: 4562-132, 4562-143, ~~4362-144~~ 4699-40

AJ 11-19-15

Testing at the lower certified limit (LCL) is required for registration, no aged batch is necessary.

Product Description:

- ☐ Quaternary ammonia
- ☐ Iodophor
- ☐ Sodium hypochlorite
- ☐ Peracetic acid
- ☐ Peroxide
- ☑ Other Sodium percarbonate and TAED

Approximate Test Substance Active Concentration (upon submission to Accuratus Lab Services):

AJ 11-19-15 (17) @16.5% Sodium percarbonate and 4% TAED

(This value is used for neutralization planning only. This value is not intended to represent characterization values.)

Neutralization/Subculture Broth: ☒ Letheen Broth + 3.0% Tween 80 + 0.4% Sodium Lauryl Sulfate + 0.3% Lecithin + 3% Saponin + 0.1% Histidine + 0.5% Sodium Thiosulfate + 0.01% Catalase

(NOTE: All broth must also serve as an appropriate growth medium for the test organism)

☐ Accuratus Lab Services' Discretion. By checking, the Sponsor authorizes Accuratus Lab Services, at their discretion, to perform neutralization confirmation assays at the Sponsor's expense prior to testing to determine the most appropriate neutralizer. (See Fee Schedule).

Storage Conditions:

- ☑ Room Temperature
- ☐ 2-8°C
- ☐ Other:

Hazards:

- ☐ None known: Use Standard Precautions
- ☑ Material Safety Data Sheet, Attached for each product
- ☐ As Follows:

Product Preparation

- ☐ No dilution required, Use as received (RTU)
- ☑ *Dilution(s) to be tested:

62.4g/ 1 liter (example: 1 oz/gallon) defined as 1 pre-weighed packet (amount of test substance) + 1 Liter (amount of diluent)

- ☐ Deionized Water (Filter or Autoclave Sterilized)
- ☐ Tap Water (Filter or Autoclave Sterilized) - All tap water is softened; the water hardness for the batch of tap water used will be determined and reported.
- ☑ AOAC Synthetic Hard Water: 100 PPM
- ☑ Other See modifications for preparation instructions

***Note: An equivalent dilution may be made unless otherwise requested by the Sponsor.**

Test Organism(s): ☑ *Staphylococcus aureus* (ATCC 6538)
☑ *Klebsiella pneumoniae* (ATCC 4352)

Carrier Number: 5 test carriers per batch and 3 population control carriers

Fabric Carrier Type: ☑ 100% Plain Cotton Weave ☐ Other:
☑ 100% Polyester

Exposure Time: 4.5 minutes **Exposure Temperature:** Room temperature (18-25 °C)

Organic Soil Load:

- ☑ Minimum 5% Organic Soil Load (Fetal Bovine Serum)
- ☐ No Organic Soil Load Required
- ☐ Other:

SPRAY BOTTLES USED IN TESTING

To ensure expected levels of product are delivered, it is recommended that the Sponsor provide the spray bottles used in testing. Please indicate the desired source of the sprayer bottles used in testing:

- ☐ Sprayer(s) and bottle(s) are provided by the Sponsor
- ☑ General purpose spray bottle(s) are to be provided by Accuratus Lab Services
- ☐ The spray nozzle(s) are provided by the Sponsor and general purpose bottle(s) will be provided by Accuratus Lab Services

AJ 11-19-15

TEST SUBSTANCE SHIPMENT STATUS

(This section is for informational purposes only.)

- ☑ Test Substance is already present at Accuratus Lab Services.
- ☑ Test Substance has been or will be shipped to Accuratus Lab Services.
  Date of expected receipt at Accuratus Lab Services: Lot 4699-40 arrives 11-20-15
- ☐ Test Substance to be hand-delivered (must arrive by noon at least one day prior to testing or other arrangements made with the Study director).

REGULATORY AGENCY(S) THAT MAY REVIEW DATA

- ☑ U.S. EPA
- ☐ Health Canada
- ☐ Therapeutic Goods Administration (Australian TGA)

COMPLIANCE

Study to be performed under EPA Good Laboratory Practice regulations (40 CFR Part 160) and in accordance to standard operating procedures.

- ☑ Yes
- ☐ No (Non-GLP or Development Study)

PROTOCOL MODIFICATIONS

- ☐ Approved without modification
- ☑ Approved with modification

A draft report will be provided for review prior to finalization. Per Sponsor request, the test culture will be standardized to target 5.88 – 6.88 $\log_{10}$ CFU/carrier. The test substance is provided in 62.4gm doses. AJ 11-19-15

For Preparing Test Substance:

1. Prepare 100 ppm hard water. Temperature of water should be 25°C ± 5°C.
2. Stir 1 L of 100 ppm AOAC hard water with a magnetic stirrer in order to produce about a 1 inch deep vortex.
3. Slowly add 1 dose (62.4g) of powder to the 1 L of water. This can best be done by gently tapping the powder vessel and sprinkling the powder into the water just off the center of the vortex.
4. Stir the mixture until all powder is dispersed but no longer than 20 minutes for the 62.4 g/1L mixture.
5. Please record the needed dissolution time. Record the time needed to achieve full dispersion.
6. The test solution must be used within 40 minutes following mixing (i.e. 60 minutes from when powder is added to water).

PROTOCOL ATTACHMENTS

Supplemental Information Form Attached - ☐ Yes ☑ No

TEST SUBSTANCE CHARACTERIZATION & STABILITY TESTING
[Verification required per 40 CFR Part 160 Subpart B (160.31(d))]

☐ Characterization/Stability testing is not required (For Non-GLP or Development testing only)

OR

Physical and Chemical Characterization (Identity, purity, strength, solubility, as applicable) of the test lots

☑ **Physical & Chemical Characterization has been or will be completed prior to efficacy testing.**

GLP compliance status of physical & chemical characterization testing:
☑ Testing was or will be performed following 40 CFR Part 160 GLP regulations
☐ Characterization has not been or will not be performed following GLP regulations

Check and complete the following that apply:
☑ A Certificate of Analysis (C of A) may be provided for each lot of test substance. If provided, the C of A will be appended to the report.
☑ Testing has been or will be conducted at Accuratus Lab Services under protocol or study #: _____
☐ Test has been or will be conducted by another facility under protocol or study #: _____

☐ **Physical & Chemical Characterization was not or will not be performed prior to efficacy testing.**

Stability Testing of the formulation

☑ **Stability testing has been or will be completed prior to or concurrent with efficacy testing.**

GLP compliance status of stability testing:
(GLP compliance is required by 40 CFR Part 160)
☑ Testing was or will be performed following 40 CFR Part 160 GLP regulations
☐ Stability testing has not been or will not be performed following GLP regulations

Check and complete the following that apply:
☐ Testing has been or will be conducted at Accuratus Lab Services under protocol or study #: _____
☐ Test has been or will be conducted by another facility under protocol or study #: _____

☐ **Stability testing was not or will not be performed prior to or concurrent with efficacy testing.**

If test substance characterization or stability testing information is not provided or is not performed following GLP regulations, this will be indicated in the GLP compliance statement of the final report.

APPROVAL SIGNATURES

SPONSOR:

NAME: Ms. Rhonda Jones TITLE: Agent

SIGNATURE: [signature] DATE: 11-19-15

PHONE: (260) 244 - 6270 FAX: (260) 244 - 6273 EMAIL: rjones@srcconsultants.com

For confidentiality purposes, study information will be released only to the sponsor/representative signing the protocol (above) unless other individuals are specifically authorized in writing to receive study information.
Other individuals authorized to receive information regarding this study: ☐ See Attached

Steven (Sam) Adamy, SRC Staff

Accuratus Lab Services:

NAME: Maggie Brusky Study Director

SIGNATURE: Maggie A Brusky Study Director DATE: 11/23/15